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## STUDIES ON ILLINOIS CERCARIAE \*

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For two and a half years the writer has been studying the cercariae of Illinois. Material has been secured from the Rock River drainage at DeKalb and Mt. Morris, and from the Sangamon drainage at Urbana and Homer. In all cases the larvae were found in the common snails of the area, *Planorbis trivolvis*, *Physa gyrina* and *Gonio-basis pulchella*.

The snails were dissected and the infected organs teased out in one-half normal saline solution. This solution was found to be quite satisfactory as an isotonic medium, although different species of fluke larvae vary considerably in their osmotic equilibria. In all cases in the present paper the exact course of the excretory capillaries has been traced and in six species the minute structure of the flame cells has been studied. A portion of each infected tissue has been preserved. Gilson's fluid has been used as a fixing agent. Toto mounts of the flukes and sections of the infected tissues have been studied to supplement the work on the living material. The former have been stained in a dye consisting of one part each of stock Delafield's hematoxylin and Ehrlich's acid hematoxylin in twenty-five parts of an aqueous solution of saturated ammonium alum. Sections have been stained with Delafield's hematoxylin with an eosin counterstain. The preserved material has been most valuable in observing the genital organs and the histological structure of the worm.

In a previous study the writer (1918) has shown that the infection of the mollusks of the Bitter Root Valley, Montana, varies within very limited areas but that the same larvae are found in the valley from one season to another. The Illinois cercariae are much more variable both in seasonal and locational distribution, but are on the whole more cosmopolitan species. *Cercaria trivolvis* and *C. isocotylea*, described by Cort for Urbana in 1913, were found by the writer in the Normal School pond at DeKalb in August, 1917. A furcocercous form, *Cercaria gigas*, secured from *Planorbis trivolvis* at DeKalb and

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from *Physa gyrina* in Pine Creek of Rock River in August, 1917, was not present in the DeKalb mollusca in October, 1917, but was found in *Planorbis trivolvis* at Urbana during this month, although not previously reported from that area. On the other hand, the finding of certain monostome cercariae (*Cercaria robusta* and *C. aurita*) only in a single locality does not necessarily argue for the limitation of these species to that area alone.

Habits and morphologic features of cercariae have been studied in isolated instances. Significant investigations of recent years have been made by Ssinitzin (1905, 1911), Cort (1915), and Faust (1918). Of the fifteen species described by Cort, ten were found within the state of Illinois. Of these, four were taken from Urbana, four from Chicago, and one each from Rockford and Mahomet. One species (*C. inhabilis*) was found in *Planorbis trivolvis* at Urbana and also at Lawrence, Kansas, (Cort, 1915), one (*C. douthitti*) at Chicago and Douglas Lake, Michigan, (Cort, 1917), and another (*C. diastrophia*) at Chicago and Lawrence, Kansas (O'Roke, 1917). The present study includes records of two species (*C. isocotylea* Cort and *C. trivolvis* Cort) from both Urbana and DeKalb, and *C. gigas* from Urbana, DeKalb and Pine Creek.

#### MONOSTOME CERCARIAE

##### *Cercaria robusta* nov. spec. (Figs. 1-5)

Host: *Physa gyrina*.

Locality: Normal School pond, DeKalb.

Collected in August and October, 1917.

Parthenita: redia.

The worm for which the name *Cercaria robusta* is proposed is broadly spatulate, more or less acutely ovoid anteriorly and obtusely rounded posteriorly. The tail is extremely muscular, hence capable of great contraction, and is much shorter than the body. The length of an average mature specimen is 0.32 mm. and the width is 0.15 mm. The tail is about 0.15 mm. long and 0.06 mm. wide at its proximal end. There is a pair of lateral eye-spots and a single median eye on the dorsal surface just behind the pharynx. Melanoidin granules are imbedded in the hypodermal tissues over the central nervous system and extend posteriorly along six lines, two dorsal, two lateral, and two ventral, marking out superficially the main posterior nerve trunks (Faust, 1918). At the postero-lateral margins of the animal are a pair of locomotor pockets, each of which is provided with a few large gland cells at its inner end (Fig. 1).

The redia averages about 2 mm. in length by about 0.4 mm. in width. Its pharynx is small but powerful. The rhabdocoel gut is longer than the cavity of the redia and is coiled forward in the region

of germ-ball proliferation. The posterior end of the redia is frequently top-shaped. The germinal epithelium lies in this posterior tip, from which the germ balls are derived. All of the rediae observed were producing cercariae. Neither lateral feet nor collar nor birth pore have been observed in the redia of this species.

The excretory system opens dorsad from an oval bladder through a small pore. Two main trunks arise from a common head just over the posterior portion of the pharynx. Each trunk receives a common external lateral halfway back from the anterior end of the system. The lateral is found to be derived from an anterior and a posterior longitudinal canal which run parallel to the main trunk. These external canals have their origin in very small capillaries. A single flame cell is probably at the head of each capillary (Fig. 1).

From an ovate pharynx  $38\mu$  in diameter the digestive tract leads back through a short esophagus to a pair of furcae which reach to the subcaudal region of the body. The furcae are not conspicuous in the living animal and are made out with difficulty in the toto mounts but may be observed in sections.

The nervous system of the animal is outlined superficially by the pattern of the melanoidin granules. The main cerebral mass is dorsal to the esophagus, forming a saddle over that organ. The pair of ventral trunks is the most conspicuous of the anterior series, while all three trunks of the posterior series, dorsals, laterals, and ventrals, are equally well developed. Transverse commissures between posterior trunks are frequent. The pair of eye-spots is lateral to the ganglion center on the dorsal side, just anterior to the junction of the dorsal and lateral nerves. These eye-spots arise from the posterior dorsal trunks. The median eye-spot is smaller and the granules are less definitely massed than those in the paired eye-spots. This cyclopean eye is immediately in front of the cerebral mass. The eye structure is similar to that of *Cercaria pellucida* (Faust, 1918).

The genital organs are clearly outlined in *Cercaria robusta*. The ovarian cell mass is skull-cap shaped; it lies just in front of the excretory bladder. A uterine duct is represented by a chain of cells which arises just anterior to the ovary and ends in the anterior third of the worm, a short distance behind the lateral eyes. A small cell mass at its anterior end is the vagina. The two small testes are to the right and left of the excretory bladder. The vasa efferentia arise from these cell masses and, bending around the ovary, unite just anterior to this organ to form the vas deferens. The vas deferens lies to the left of the uterus and parallels it to the region of the vagina, where it ends in a small swollen mass, the cirrus. The vitelline glands are aciculate in outline. They are composed of three pairs of glands

in the outer series and four pairs and a double median gland in the inner series. The median gland is the anteriormost of the inner series and represents a fused pair. Aside from this single modification the glands are similar in position and number to those described by the writer for *Cercaria pellucida*, *C. konadensis*, and *C. urbanensis*.

The conspicuous structures of the living *C. robusta* are the longitudinal muscle fibers of the tail. More deeply located in this organ are six pairs of large gland cells. These correspond in grouping to the six pairs of gland cell groups in the tail of *C. konadensis* and are identical in number and structure to the six pairs of gland cells in the tail of *C. urbanensis*.

The mature *Cercaria robusta* breaks through the wall of the redia and penetrates the liver tissue of the host. It may either work its way to the free water or encyst in the liver sinuses. The movement accomplished by the coordination of oral sucker and posterior locomotor pockets is slight; most of the locomotion comes from the activity of the tail. Due to its extensive muscularization, this organ acts as a powerful whip-lash, stirring up a whirlpool eddy all around it by its violent movements. When this cercaria is set free into the water it attaches itself by the oral sucker, while a whirlpool movement of the entire worm is initiated by the tail. Encystment starts immediately. Beginning at the oral sucker it proceeds rapidly backward, limiting the size of the whirlpool as encystment continues. Finally a cyst has been formed around the entire worm, while the free tail, attached to the cyst only by a fibril, continues its characteristic movement. Then the worm within the cyst twists around and loosens its connection with the fibril. For a while the tail keeps lashing after all connection with the encysted worm has been broken, but the movement of the organ tends to become less violent and at length ceases entirely.

*Cercaria aurita* nov. spec. (Figs. 6-8)

Host: *Goniobasis pulchella* (Anthony).

Locality: Salt Fork of Sangamon River, Homer.

Collected: October, 1917.

Parthenita: redia.

This species is designated as *Cercaria aurita* because of the lappet processes which characterize the worm just lateral to the pair of eyespots. The animal has a length of 0.57 mm. and a width of 0.19 mm. The tail is 0.33 mm. long and 0.08 mm. wide at the base. When the animal elongates the sides are parallel and the animal is roughly rectangular, save for a blunt rostrum in front of the auricular prominences. At the postero-lateral angles are a pair of locomotor pockets, which are distinctly helpful in locomotion. When the body contracts

it becomes pear-shaped. The tail is comparatively useless. The body has a dirty grayish-brown appearance.

*Cercaria aurita* develops within a redia about 1.5 mm. long and 0.4 mm. in diameter. The pharynx is small and leads into a large rhabdocoel gut which extends through the body cavity about three fourths the way to the posterior end. Only a few cercariae develop at any one time within the redia. They break through the heavy wall of the redia and worm their way through the water.

The excretory tract in *C. aurita* is more primitive than that of any previously described monostome cercaria. A small oval bladder at the caudal end of the body receives two dilated trunks through a common reservoir. The trunks are very short and become reduced to the size of capillaries at the loci where they turn forward. These capillaries can be traced forward for only a short distance. This reduction in the excretory system constitutes a remarkable differentiation from the circuitous system in previously described monostome cercariae. It is further distinguished by the absence of excretory granules in the canals and trunks.

The digestive tract is prominent and easily seen in the living worm. A subspherical pharynx,  $13\mu$  in diameter, leads into a long esophagus that extends through somewhat more than the anterior third of the worm. The furcae are of a length equal to the esophagus. At their blind end they are distended, so that they appear club-shaped. The cells lining the digestive tract are large and glandular.

The nervous center of the worm is diffuse. It covers considerably more ground than that of *C. robusta*. This fact is superficially recognized by the diffuse arrangement of the melanoidin granules just beneath the basal membrane of the animal. No anterior nerve trunks are prominent; all three pairs of posterior trunks are easily made out although their transverse commissures are inconspicuous. The pair of lateral eye-spots on the dorsal side is well developed. These eyes are set out some distance from the center of the body. Their connection with the brain ganglia has not been studied.

The genital organs are most unusual in their limited extent. Except for vitelline elements they are confined to the middle third of the body. The ovary is represented by a small irregular mass of cells median in position. A short string of cells, the uterine cells, leads to a spherical mass a short distance anteriorly, the vagina. The testes are slightly behind the ovary, rather irregular in appearance, and appreciably larger than the ovary. Their vasa efferentia coalesce in front of the ovary to the left of the uterus, and the common vas deferens runs forward to a spherical cirrus pouch. The vitelline glands could not be definitely made out. They are diffuse in their structure. A

small anterior portion occurs just in front of the testes. A large branch extends posteriad on the ventral side of the worm. This pattern of vitelline distribution has not been reported thus far for monostome cercariae and differs from the vitelline structure of other monostome larvae as markedly as the other features of this worm differ from those of described monostome cercariae.

The cercaria is not conspicuously active. It comes out of the mature redia as a sluggish, crawling worm. The tail is a hindrance rather than an aid in movement, for it is dragged along behind the body without any independent movement. The posterior locomotor pockets cooperate with the oral sucker in the attachment of the worm to the crawling surface. These pockets are not muscular but are provided with several large gland cells.

Encystment has not been observed, although the large number of semi-opaque cystogenous glands must function in the secretion of a cyst. The worm soon disintegrates when placed in a hypotonic medium.

#### ECHINOSTOME LARVAE

##### *Cercaria chisolinata* nov. spec. (Figs. 9-13)

Host: *Physa gyrina*.

Locality: Pine Creek of Rock River, near Mt. Morris.

Collected: August, 1917.

Parthenita: redia.

This echinostome cercaria is named *Cercaria chisolinata* because of the crossing of the excretory tubules at the anterior end of each lateral system. The worm is 0.3 mm. long and one-third as wide. The tail is about 0.5 mm. long and 0.09 mm. wide at the proximal end. At the anterior end the collar prominence is provided with about forty small sharp spines consisting of a series in two alternating rows on the dorsal side of the worm and extending ventrad to a region just below the anterior end of the excretory system. Each spine measures about  $20\mu$  in length. The oral sucker is  $44\mu$  in diameter. A ventral sucker of equal diameter is situated a short distance behind the middle of the body. The tail is marked by longitudinal muscle fibers, which become less conspicuous distad.

The redia is a large parthenita, 1.5 mm. long and 0.45 mm. in section across the feet. These appendages occur one-third the distance from the posterior end of the worm. A collar prominence is found near the anterior end. Just behind it on the ventral side is the conspicuous birth pore. The wall of the redia is thick. At the anterior end there is a pharynx  $10\mu$  in diameter. Behind this there is a dwarf rhabdocoel gut, barely twice the length of the pharynx. Many cercariae develop within the redia at the same time. The larvae seem to develop in batches, so that the production tends to be rhythmical

The excretory system is typically echinostome in character. The bladder receives the pair of lateral trunks through a common chamber. These trunks are most dilated just anterior to the acetabulum, where they are filled with granules. Just behind the pharynx they narrow down to the dimensions of capillaries. At the posterior margin of the oral sucker each capillary bends abruptly outward, then backward, crossing back on itself at the junction of trunk and capillary. Here it ends in a flame cell. One flame cell is also found in the outward bend and one at the reflexing of the capillary. Three flame cells such as these at the anterior end of the system are probably found in the majority of echinostome larvae. A single median tubule in the tail cares for excretory wastes in that organ.

The digestive tract is surrounded by a very small pharynx at its anterior end. The esophagus continues to the anterior margin of the acetabulum. Here the ceca arise and continue to the caudal extremity of the worm. There are two series of mucin glands, with many members to each series. They empty through common ducts into the oral atrium of the cercaria.

The genital organs are poorly developed. An ovarian cell mass is found just anterior to the bladder; another cell mass is found at the antero-lateral margin of the acetabulum. Fine lines of vitelline follicles are traceable anteriorly and posteriorly.

The animal is filled with small cystogenous glands, which arise late in the development of the larva through differentiation of indifferent parenchyma cells. The worm normally decaudates and encysts in the host tissue. The cyst wall is thick and firm, providing a safe abode for the worm until a transfer of hosts is effected.

*Cercaria acanthostoma* nov. spec. (Figs. 14-17)

Host: *Planorbis trivolvis* and *Physa gyrina*.

Locality: Urbana.

Collected: October 29, 1917.

Parthenita: redia.

This echinostome larva resembles the typical larvae of the group in having a very muscular body and an active tail, in the possession of a collar prominence with numerous spines, and in an abundant supply of cystogenous material. The body is 0.3 mm. long and 0.12 mm. wide, while the tail is 0.44 mm. long and 0.044 mm. wide at the proximal end. The oral sucker is  $58\mu$  in diameter and the ventral sucker is  $65\mu$  in diameter. A unique character of this larva is the group of six spines inserted in a single row in the roof of the oral sucker, with points directed forward. The larva is named *Cercaria acanthostoma* because of this oral spine group. The ordinary spines around the collar number from thirty-four to thirty-eight. They are sharp-



pointed and inserted in an irregular row. The tail is crenate along the margin. Although this feature is probably due to the muscular elements of the caudal organ, the wavy outline of the organ is a constant character.

The redia is comparatively small, 0.3 mm. long and 0.058 mm. in diameter. The feet are situated three-fifths the distance from the anterior end. A collar prominence is found a little way from the oral end. The pharynx is small and leads into an inconspicuous thick-walled rhabdocoel gut. The embryos develop at the posterior end of the parthenita. They press forward as they develop.

The excretory system in the body of *Cercaria acanthostoma* consists exclusively of a bladder and delicate tubules and capillaries. The bladder is biconvex with muscular walls. The pair of main tubules enters the bladder from the extreme lateral margins. Along the course of each tubule are thirteen flame cells, eight on the inner margin of the tubule and five external to the tubule (Fig. 14). At the extreme anterior end of the excretory system the tubule reflexes and then fuses to form a delta, at each angle of which there is one flame cell. Thus the total number of flame cells along the entire course of the tubule is sixteen. This system is much more delicate than the more common echinostome type with the large lateral trunks and capillary system. The excretory system in the tail is confined to a long sac-like reservoir extending the entire length of the organ without any definite wall or lining. Near the proximal end it frequently bulges out on each side to form a lateral reservoir. No flame cells were distinguishable in the tail.

The digestive tract has a prepharynx region, a large pharynx  $32\mu$  in diameter, an esophagus extending to the anterior margin of the acetabulum, and ceca extending as far posteriad as the excretory bladder. Mucin glands are present in a biserial arrangement.

The nervous system is well developed. The six main posterior trunks are visible in the toto mounts. The pair of posterior ventrals is unusually large.

The genital cell masses consist of a pyriform ovary and a vagina along the midline just in front of the acetabulum.

The animal is filled with numerous cystogenous glands, yet encystment is slow and infrequent. When it does occur the cyst membrane which is formed is thin and tough. Through it the spines and excretory system are visible.

*Cercaria trivolvis* Cort

Host: *Planorbis trivolvis*.

Locality: Normal School pond, DeKalb; drainage ditch, Urbana.

Collected: November, 1916; August, 1917.

Parthenita: redia.

This species was first described by Cort in 1914 and again in 1915. The writer has been able to examine the material from the same host in the same and in a different locality. Certain points of structure not described in the original accounts have been observed.

The specimens studied by the writer were somewhat smaller than those described by Cort. They averaged in body length 0.34 mm., while Cort's specimens had an average length of 0.38 mm. Their body width was 0.11 mm. as contrasted with Cort's width measurement of 0.12 mm. Likewise the tails of the writer's specimens were about 0.44 mm. as compared with 0.5 mm. in the individuals worked over by Cort. On the other hand the oral sucker and acetabulum of the DeKalb specimens measured  $50\mu$  and  $58\mu$ , respectively, as compared with  $43\mu$  and  $49\mu$  of Cort's material. This difference in size may be entirely dependent on the degree of maturity or on nourishment, while the size of the suckers may depend largely on the degree of expansion or contraction.

In addition to the characters described by Cort, the writer has found paired right and left clumps of salivary-mucin glands filling practically the entire body from pharynx to acetabulum. Each cluster is oval and consists of a very large number of minute cells with large dilated nuclei. These gland cells are similar to those described by the writer for *Cercaria reflexa* (1918, Fig. 134). They differ, however, in arrangement and distribution.

The crevices between the body wall and the salivary-mucin glands are packed with cystogenous glands, as is also the greater portion of the posterior portion of the body. The individual gland cell is polygonal and contains several elongate cyst granules. These granules may be compared to concentrated gelatin tablets, capable of enormous swelling by water inclusion, when the stimulus for cyst formation is at hand. Then by a rapid wriggling the tail is thrown off and a thick gelatinous cyst is secreted around the larva. Through this cyst none of the organs can be definitely made out. Only the large excretory granules are apparent through the cyst membrane.

#### STYLET LARVAE

##### *Cercaria stilifera* nov. spec. (Figs. 18, 19)

Host: *Physa gyrina*.

Locality: Pine Creek of Rock River, near Mt. Morris.

Collected: August, 1917.

Parthenita: sporocyst.

This cercaria is large for a stylet larva, 0.32 mm. in length, 0.17 mm. wide in the region of the acetabulum, and possesses a tail 0.23 mm. long and 0.07 mm. wide at the proximal end. There is a pair of caudal pockets at the junction of body and tail, with a small number

of sharp spines directed mesad. The oral sucker is large, about  $85\mu$  in diameter, while the acetabulum, slightly posterior to the middle of the body, is only  $58\mu$  in diameter. The quill inserted in the dorsal wall of the oral sucker is a simple structure,  $32\mu$  long, and reinforced only at the base.

The cercaria develops in a sporocyst of irregular contour, about three or four times as long as the transverse diameter of the parthenita. The shape of the sporocyst is largely dependent on the movement of the cercariae within the sac. The wall of the sporocyst is extremely thin and delicate and is ruptured with the slightest pressure. Immature cercariae will not live in half-saline solution.

The excretory system in the wall of *Cercaria stilifera* consists of a bladder slightly muscular, which changes in shape from oval to squarish in surface view, a single narrow reservoir directly anterior to the bladder, a pair of lateral cornua, and two main body tubules. One of these tubules is directed posteriad, the other runs anteriad just lateral to the acetabulum and at the anterior half of the worm forms two branches, the inner one of which ends in the region of the pharynx and the outer one of which ends over the oral sucker. All of these tubules have several tributaries, each one of which originates from a pair of capillaries. At the inner end of each capillary there is a flame cell. The excretory system in the tail consists of a long awl-shaped reservoir at the proximal end immediately behind the bladder and a single median tubule with some ten tributaries, each of which arises from the junction of the two capillaries.

The digestive tract is composed of a short, narrow prepharynx, a small pharynx  $17\mu$  in diameter, a long esophagus extending almost to the acetabulum, and a pair of short ceca which barely suggest the bifurcate nature of the system. Opening into the oral atrium at the extreme sides of the large sucker are the two groups of salivary-mucin glands. There are about twelve glands in each lateral group, each gland having an individual duct to the subatrial region. Here they all empty into an enlarged portion of the system filled with granules. A single duct connects this dilation with the oral atrium.

The genital organs are represented in *C. stilifera* by a knobbed mass of cells posterior to the acetabulum, a thick cord of cells on the left of the embryo just within the limits of the acetabulum, and a thick tubule just under the anterior half of the acetabulum. All of these are joined together in the order named. In addition conspicuous masses of vitelline masses are found at the sides of the body in club-shaped aggregates extending forward to the pharynx and posteriad to the bladder. The germ cells in the posterior part of the body lie ventrad to the excretory cornua; the cell mass more anteriad lies above the ventral sucker.

The nervous system is somewhat degenerate. In the region of the pharynx is a large mass of diffuse fibers, which constitute the ganglion mass. The nerve trunks are not easily distinguished.

Cystogenous glands are found scattered through the body. They are large and relatively few in number. Their nuclei are oval to spherical, and the granules in the cytoplasm are acidophilic, as contrasted to the basophilic cystogenous granules of other species. This gland structure is probably closely correlated with the slow decaudation of the animal and infrequent encystment, and suggests that the worm gains entrance to the next host either through active swimming or the next host eating the larva while it is yet within its primary host.

*Cercaria isocotylea* Cort (Figs. 20-24)

Host: *Planorbis trivolvis*.

Locality: drainage ditch, Urbana, and Normal School pond, DeKalb.

Collected: 1916-1917.

Parthenita: sporocyst.

This cercaria was originally described by Cort in 1914 and again in 1915. The writer's measurements for the species are far in excess of the original description, ranging from 0.2 to 0.32 mm. for body length and 0.1 to 0.12 mm. for body width, while the oral and ventral suckers both measure about 50 $\mu$ . The fact that Cort's specimens showed no cystogenous glands, together with their smaller size, suggests their immaturity. The tail in all of the specimens which the writer observed was small and not particularly active.

The stylet is described by Cort (1915: 54) as "sharp-pointed and has a thickening two-thirds of the distance from its base to its tip." The sharp, spinose portion of the stylet takes up the anterior third of the organ. The rest of the quill is set off by a nodular reinforcement on the dorsal side and is thickened dorsad toward the base (Figs. 22-24). From the base four tongue bars extend out anteriad.

At the posterior end of the worm the caudal pockets include the proximal portion of the tail. Each member of this pair is provided with a small group of spines projecting inward. The absence of these spines in Cort's description is an additional point in favor of the view that his specimens were immature. The small body spines which extend over the anterior two-thirds of the animal decrease in size from the anterior tip caudad.

Figure 20 shows the distribution of the excretory tubules in the species. At the end of each capillary is a minute flame cell. Twenty-two flame cells have been counted on each side of the body. A single unbranched tube is found in the tail.

The digestive canal has been described by Cort as undeveloped save for oral sucker, short prepharynx, and pharynx. The writer has

found a large group of gland cells in this species directly behind the pharynx. Further study has shown these cells to open into two short ceca and a miniature esophagus. These organs are at times so far dorsad that they are not in the same focus as the pharynx. The salivary-mucin glands (stylet glands of Cort) are regarded by the writer as a part of the digestive system. Their presence in echinostome and schistosome larvae, where no stylet is present, and the chemical nature of their content, make the theory of their salivary nature altogether probable. The writer has counted nine of these gland cells in each lateral group.

The genital cell masses are found dorsal to the acetabulum. On the posterior margin are found three masses, a median oval organ, the ovary, and two lateral lobed organs, the testes. Two masses dorsal to the anterior margin of the acetabulum correspond to the vagina and the cirrus sac. Stretches of vitelline glands occupy the sides of the body from the region of the pharynx to the extreme posterior margin of the body.

The cystogenous glands are similar to those of *C. stilifera*, few and vesicular. The granules are small. Decaudation occurs seldom. Encystment has been found to take place only within the host tissue. The cyst membrane is probably secreted very slowly, in contrast to that in monostomes and some echinostomes. This cyst wall is thick and gelatinous.

The topography of the various organs in these stylet cercariae, *C. stilifera* and *C. isocotylea* Cort, suggest plagiorchiiine relationships. While the writer believes that the stylet *per se* is a very general character which may be found only in larval Plagiorchiiidae or may, on the other hand, be found in other related Distomata, the opening of the genital pore anterior to the acetabulum is a more specific feature of these forms to which attention is directed. The stylet larvae previously described by the writer (Faust, 1918) have been found to possess cirrus or vaginal cell masses which open into a genital atrium anterior to the acetabulum. Magath (1918) has recently shown that a stylet cercariae in *Planorbis trivolvis* at Fairport, Iowa, develops into a worm related to the Plagiorchiiidae but differing from the Plagiorchiiidae in having a lateral genital pore. For this he has proposed the new subfamily Lissorchiinae. Unfortunately he was unable to make out any of the genital complex in this cercariae except the ovarian cell mass, so that it can not be compared item for item with those described by the writer. However, among other things, they possess in common (1) a stylet set in the dorsal wall of the oral sucker, (2) paired mucin glands, and (3) sporocyst parthenitae. A striking difference between the excretory system of the cercaria of *Lissorchis fairporti* and the plagiorchiiine cercariae is the flexing back

of the main anterior tubule in the former species and the absence of such flexing in the latter larvae. On the whole the larvae of these two families show marked relationships.

## SCHISTOSOME LARVAE

*Cercaria gigas* nov. spec. (Figs. 25-30)

Host: *Planorbis trivolvis*, *Physa gyrina*.

Locality: Normal School pond, DeKalb; Pine Creek of Rock River, Mt. Morris; drainage ditch, Urbana.

Collected: August-November, 1917.

Parthenita: sporocyst.

This larva is a giant among schistosome larvae. Its body length is 0.28 mm., its width 0.09 mm.; the unforked portion of the tail is 0.32 mm. long, and the tail furci, 0.18 mm. Iturbe and Gonzalez (1917) have stated the measurement of the cercaria of *Schistosoma mansoni* to be as follows: body length, 0.1-0.13 mm.; breadth, 0.04-0.05 mm.; unforked tail, 0.14-0.15 mm.; furci, 0.04-0.05 mm. The writer's measurements on some of Iturbe's material shows a slight excess in all of these measurements. *Cercaria douthitti* (Cort, 1915) has a body length of 0.19 mm., and an unforked tail length of 0.22 mm., while the furci measure 0.089 mm. *Cercaria tuberistoma* (Faust, 1918) has a body length of 0.2 mm., and a combined tail length of 0.32 mm. O'Roke's *C. echinocauda* has a body slightly larger and a tail nearly twice as long (1917).

*Cercaria gigas* is characterized by a pair of pigment eyes on the dorsal side, about two-fifths the body distance from the anterior end; by a small ventral sucker,  $26\mu$  in diameter; by a long unforked portion of the tail which is muscular to an extraordinary degree, and by fluted borders to the furcae. On account of a pronounced flexure at the juncture of body and tail, the animal is more often seen on the side than on the dorsal or ventral surface. In this attitude it has a characteristic irregular appearance more easily pictured than described (Figs. 28, 29). The oral sucker sticks out anteriorly like a snout. It is covered with minute spines and is invertible. The acetabulum protrudes some distance ventrad. Behind it a group of gland cells bulges ventrad. These glands are easily made out in the living animal as an oval mass of yellowish-white in the midst of a grayish background.

Often the ventral surface is streaked with pigment, especially just behind the eye-spots. Melanoidin granules are frequently distributed over both dorsal and ventral surfaces.

The unforked portion of the tail is large and powerful. Many longitudinal muscle fibers run the entire length of the organ. These are reduced in size and number in the furcae, which are thin and paddle-like with their edges directed dorsoventrad.

The sporocysts are long, irregular sacs, most usually pointed posterioriad and muscular in the anteriormost portion. The walls of the sporocyst are moderately thick. The cercariae appear to develop in batches.

The excretory organs have been worked out in detail in *Cercaria gigas*. The bladder is small and oval. Two main tubes enter into it side by side at its anterior end. These reach forward to the region of the eye-spots. Along the course of each tube are ten flame cells. A main tube in the unforked portion of the tail receives a tubule from each furca. At the proximal end of the caudal organ a lateral tubule on each side flows into the main tube. At the head of each lateral tube is a flame cell. The tube splits and reunites just before it enters the bladder.

*Cercaria gigas* has no pharynx. Esophagus and ceca are also wanting. Very large ducts with thick walls empty into the sides of the oral sucker. These ducts are the openings of two paired groups of gland cells. The anterior of these groups consists of several gland cells centering around the acetabulum. The protoplasm of these cells is granular and the nuclei are small. The posterior group consists of many cells, small and chromophilic. All of these glands are salivary-mucin in character. Their large number is unique among Schistosomatid larvae.

The nerve tracts are well defined. Anteriad there are three main pairs of trunks. Posterioriad the laterals are lacking and the dorsals soon fuse with the ventrals. The eye-spots have a direct connection with the anterior dorsal trunks.

Only one group of genital cells is found, the testes mass, just behind the acetabulum.

The cercaria does not encyst. It probably reaches its definitive host by direct method and bores its way through the tissues to the blood stream.

By gross inspection *C. gigas* is likely to be confused with *C. echinocauda*. This resemblance of the two species is pronounced, save for the longer tail stem in *C. echinocauda*. Possible confusion of these two forms warrants a discussion of their similarities and differences. The writer has been fortunate to secure material from O'Roke and has therefore been able to check up the items from the material itself. *C. echinocauda* is longer and wider; its tail length is disproportionally greater. The furcae of both species have about the same measurement. But while the furcal fins of *C. gigas* are closely fluted, those of *C. echinocauda* are flat and braced with radial thickenings so that they were mistaken by O'Roke for spines. The longitudinal muscles in the tail stem of both species are prominent, but they are coarser in *C. gigas*.

In *C. echinocauda* the furcae arise slightly lateral, with a stub of the tail stem extending slightly distad; in *C. gigas* the furcae arise from a common center in the midline. O'Roke (1917:171) mentions the flexure at the junction of body and tail of *C. echinocauda*. This flexure is much more pronounced in *C. gigas*, so that it is difficult to get a frontal mount.

*C. echinocauda* has no spines at the oral end of the body such as are found on *C. gigas*. The pigment eyes of *C. echinocauda* are cup-shaped, with the opening dorsolateral; the eye-spots of *C. gigas* are long, sac-shaped organs, with the long axis extending dorsoventrad. No pigmentation other than that of the eyes has been observed by the writer in *C. echinocauda*.

Internally the structural differences of the two species are pronounced. The oral pocket in *C. gigas* ends blindly; there is neither pharynx nor esophageal glands. In *C. echinocauda* there are a few attenuate esophageal glands at the base of the oral pocket. The mucin-gland ducts in both species are large and conspicuous; but while there are two structurally differentiated groups of mucin glands in *C. gigas*, with many glands in each group, there are a few large glands of only one kind in *C. echinocauda*. The latter are chromophobic. The testes cell mass in *C. gigas* is composed of several small entities immediately behind the acetabulum; in *C. echinocauda* relatively few units compose this germinal mass and the gland is a considerable distance behind the acetabulum. Moreover, other genital cell masses may be made out distinctly in the region of the acetabulum of the latter species. O'Roke has not made out the excretory tubules or flame cells in the body of *C. echinocauda* so no comparison of these organs in the two species can be made.

*C. echinocauda* is described as the offspring of a redia, whereas the evidence of studies on other cercariae of the furcocercous group preponderates in favor of the development of these cercariae within sporocysts. Such sporocysts are at times muscular at the anterior end, with a pouch-like structure which serves as a sucker, but in no case has a true pharynx or rhabdocoel gut been demonstrated. The material of *C. echinocauda* examined by the writer contains no parthenitae, but the general outlines of O'Roke's figures (1917, Figs. 37, 41, 47) suggest sporocysts rather than rediae. This question must be carefully checked before it is finally settled.

*Cercaria minor* nov. spec. (Figs. 31-33)

Host: *Physa gyrina*.

Locality: Normal School pond, DeKalb.

Collected: August, 1917.

Parthenita: sporocyst.



*Cercaria minor* is much smaller than *C. gigas*. The length of the oval body is 0.14 mm. and the width, 0.068 mm. The unforked tail measures 0.2 mm., which is the same length as the furcae. The tail has a transverse diameter of  $40\mu$  at the proximal end. The oral sucker opens ventrad. It is  $23\mu$  in diameter and considerably deeper. The ventral sucker is in the posterior half of the body. It is  $26\mu$  in diameter and has a small circlet of spines within its margin. A pair of non-pigmented eye-spots is found in the region posterior to the oral sucker. Large parenchyma cells are found in the unforked portion of the tail.

The sporocyst is large and irregular, measuring up to 2.1 mm. in length by 0.27 mm. in diameter. One end is slightly muscular and is used in burrowing.

The excretory system consists of an oval bladder, flattened antieriad, and a pair of main tubules which stretch antieriad to the region of the oral sucker. Each tubule gives off two biramous inner branches and a single biramous posterior twig. The tail has a single unbranched median tubule. An eyelet anastomosis occurs between the tail and the bladder.

The pharynx is represented by a few small glandular cells. Four pairs of salivary mucin glands empty into the oral sucker thru heavy ducts.

*Cercaria minor* has not been found to encyst. It probably reaches the definitive host as a cercaria and then metamorphoses into the adult schistosome.

In an attempt to harmonize the flame cells of furcocercariae, Cort (1918) has recognized three divisions of these larvae: (1) those characterized by absence of a pharynx, tail furcae less than half the length of the tail stem, eye-spots present; (2) human schistosome larvae; (3) those with pharynx present, tail furcae almost as long as main stem. According to this grouping, *C. gigas* falls into the first class, altho it possesses ten pairs of flame cells in the body and one pair in the tail, a larger number than is found in Cort's forms, *C. douthitti* and *C. elephantis*. While *C. minor* (Figs. 31, 32) bears some resemblance to *C. douglasi*, it is much more akin to *C. gracillima* in possessing non-pigmented eye-spots and pyriform glands in the region of the esophagus which definitely denote the transformation of the pharynx region from a muscular to a glandular organ. Moreover, *C. douglasi* is classed outside of the Schistosomatidae because it has a pharynx, while *C. gracillima* has been shown to possess a definite schistosome nervous system (Faust 1918: 54). With the broadening knowledge of schistosome larvae, it seems more reasonable to recognize a complete series of larval forms from those with a pharynx sphincter (*C. douglasi*, *C. emarginatae* and perhaps *C. vivax* Sonsino), thru those with a degenerate pharynx, with or without intestinal ceca (*C. gracillima*,

*C. minor*), thru those without any pharynx, but with well developed mucin glands (*C. gigas*, *C. tuberistoma*, *C. douthitti*), to the human schistosome cercariae. For example, *C. minor*, in lacking intestinal ceca, is more closely related to *C. douthitti* than *C. gracillima*. Yet the eye-spots in *C. minor* are not pigmented.

Until the genital cell masses of each of these larvae have been carefully studied it is useless to attempt the relationships within the groups.

Thru the courtesy of Professor Henry B. Ward, the writer has been enabled to examine specimens of *Planorbis quadelupensis* Sowerly infected with schistosome larvae sent by Dr. Juan Iturbe of Caracas, Venezuela. One vial of this material (No. 17.198 Ward collection) with the accompanying label, "rediae in state of development," was found to contain no stages in the life cycle of the schistosome larva, but instead a unique tetracotyle, for which the name *Tetracotyle iturbei* is proposed.

*Tetracotyle iturbei* nov. spec.

*T. iturbei* is a pyriform fluke, 0.42 mm. in length, 0.33 mm. in width, and 0.3 mm. in thickness. The oral sucker has a diameter of  $52\mu$ , the primitive genital pore,  $42\mu$ , and the acetabulum,  $95\mu$ . Suctorial grooves, muscular in part, are located at the sides of the ceca. The worm is unarmed and is enclosed in a thin mucoid cyst. Prepharynx is lacking; the pharynx measures  $16\mu$  in trans-section; the digestive tract forks immediately behind the pharynx. The ceca extend thru the anterior half of the body. The ovary, measuring  $25\mu$  in diameter, lies midway between the acetabulum and the posterior genital pore. Vitellaria are massed into two compact chorda, which reach cephalad as far as the primitive genital pore. The ootype, dorsal to the ovary, leads posteriad thru a short duct into a small genital pouch. Large pyriform testes,  $50\mu$  in long diameter, lie lateral and somewhat anterior to the ovary. Separate efferent ducts lead into the posterior genital atrium. Anterior to the ovary is a vagina, connected by means of a coiled tube with the anterior ventral sucker, the expanded primitive genital pore. This species yields data in support of the view that the posterior, and usually degenerate, ventral sucker of holostomes is the acetabulum of distome species.

*T. iturbei* is figured (Iturbe and Gonzalez, 1917; pl. 1, figs. 1-9) as the redia of *Schistosoma mansoni*. Proof that Iturbe's "redia" is a distinct species of fluke confirms the belief that rediae are not found among Schistosomatidae.

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#### EXPLANATION OF PLATES

##### PLATE I

*Cercaria robusta*—Fig. 1.—Dorsal view, showing pigmentation, excretory system and longitudinal muscles of the tail.  $\times 170$ . Fig. 2.—Dorsal view, showing germ glands in body and parenchyma cells in tail.  $\times 170$ . Fig. 3.—Redia.  $\times 34$ . Figs. 4, 5.—Stages in encystment.  $\times 75$ .

*Cercaria aurita*—Fig. 6.—Dorsal view, showing pigmentation and excretory system.  $\times 105$ . Fig. 7.—Dorsal view, showing digestive and genital organs.  $\times 105$ . Fig. 8.—Redia.  $\times 54$ .

*Cercaria chisolena*—Fig. 9.—Dorsal view, with cystogenous glands on right of diagram.  $\times 105$ . Fig. 10.—Diagram of genital cell masses.  $\times 105$ . Fig. 11.—Dorsal view of collar spines.  $\times 170$ . Fig. 12.—Detail of anterior tip of excretory system with three flame cells.  $\times 370$ . Fig. 13.—Redia.  $\times 54$ .

##### PLATE II

*Cercaria acanthostoma*—Fig. 14.—Ventral view, showing excretory, digestive, and genital organs.  $\times 170$ . Fig. 15.—Pattern of collar spines, dorsal view.  $\times 333$ . Fig. 16.—Detail of anterior tip of excretory system.  $\times 500$ . Fig. 17.—Redia.  $\times 54$ .

*Cercaria stelifera*—Fig. 18.—Ventral view, showing excretory, digestive and genital organs.  $\times 170$ . Fig. 19.—Stylet.  $\times 370$ .

*Cercaria isocotylea*—Fig. 20.—Ventral view, showing excretory, digestive and genital organs.  $\times 170$ . Fig. 21.—Lateral view, illustrating description of spines on surface of body.  $\times 105$ . Figs. 22-24.—Dorsal, lateral and ventral views of stylet.  $\times 370$ .

*Cercaria gigas*—Fig. 25.—Dorsal view, showing eye-spots and salivary-mucin glands.  $\times 170$ . Fig. 26.—Excretory system. Fig. 27.—Sporocyst.  $\times 54$ . Figs. 27, 28.—Characteristic lateral views.  $\times 54$ . Fig. 30.—Immature cercaria.  $\times 54$ .

*Cercaria minor*—Fig. 31.—Ventral view, showing salivary-mucin glands in body and parenchyma cells in tail.  $\times 170$ . Fig. 32.—Excretory system.  $\times 333$ . Fig. 33.—Sporocyst.  $\times 34$ .

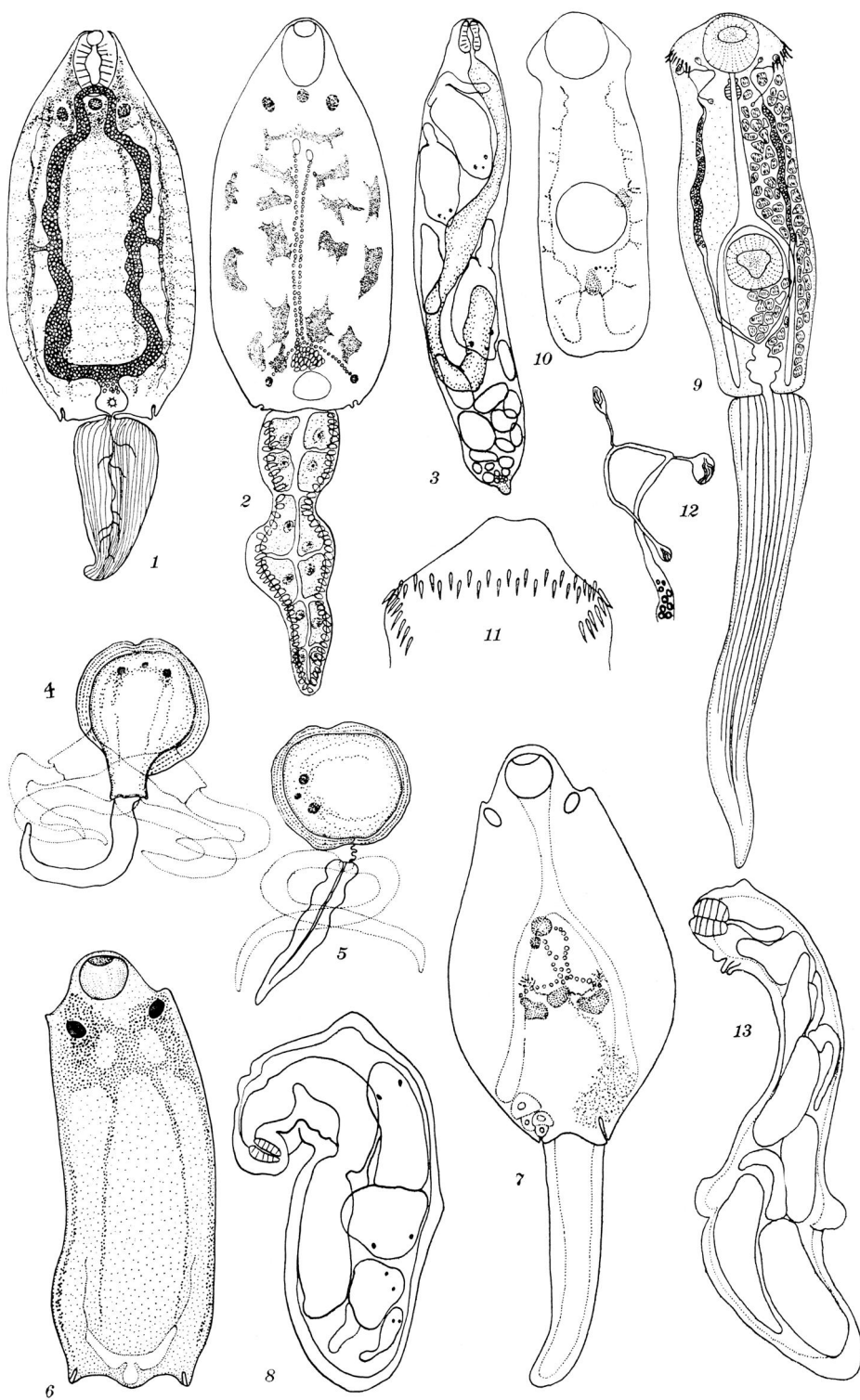


PLATE I

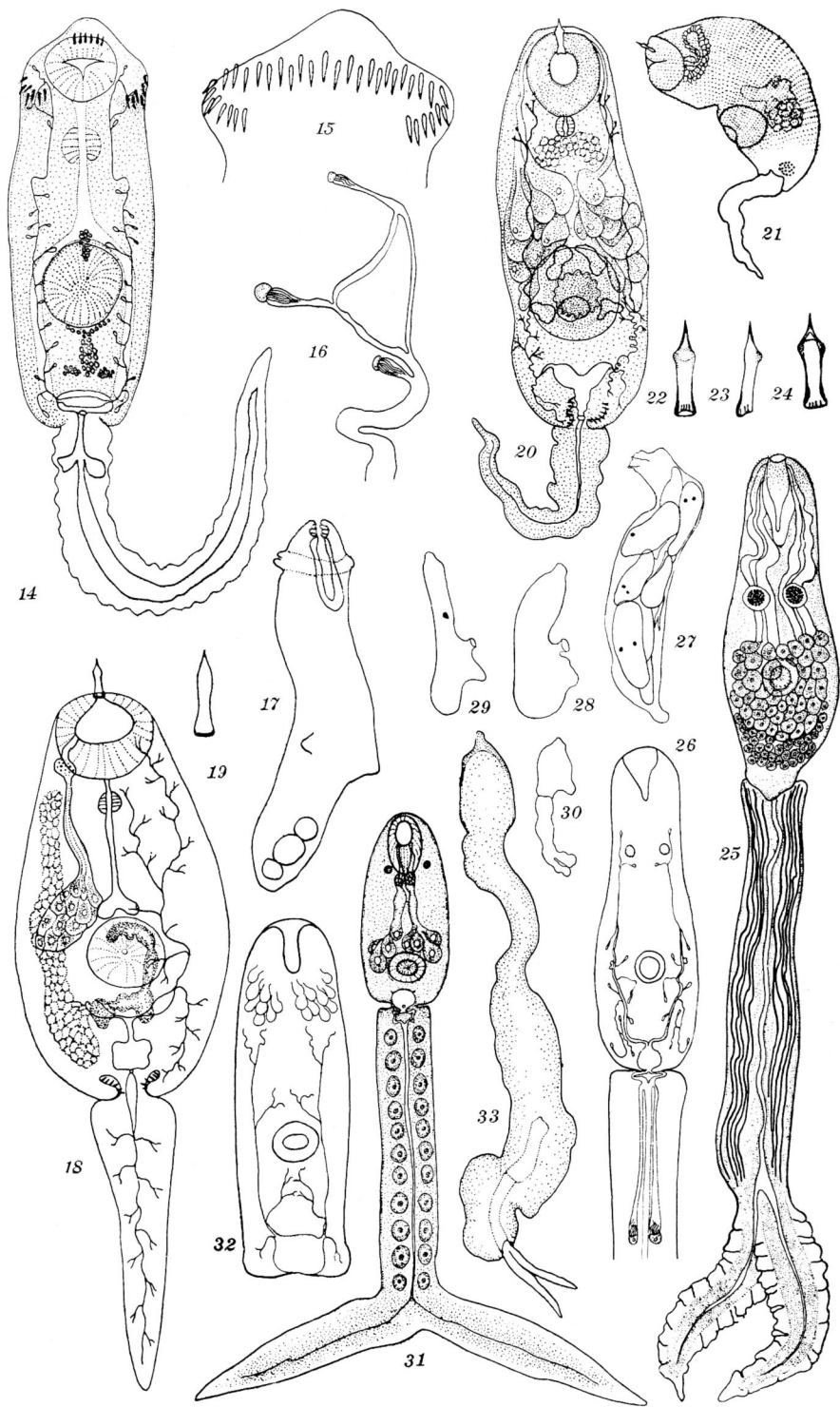


PLATE II